Integrated Water Quality and Aquatic Communities Protocol – Wadeable Streams

Standard Operating Procedure (SOP) #9: Macroinvertebrate Collection

Draft Version 1.0

Revision History Log:

Previous	Revision	Author	Changes Made	Reason for Change	New
Version	Date				Version

This SOP explains the procedures for the collection of benthic macroinvertebrates. The basic procedure used follows the EPA EMAP (Peck et al. 2006) protocol for the collection of Reach Wide Benthos (RWB). This protocol follows that of the periphyton collection in that it is not limited to the traditional targeted habitat, but rather the substrate and habitat at each of the 11 transects. Each sample from the individual transects is composited into a single sample for the entire reach.

Macroinvertebrates are collected in a D-frame net (30.5 cm wide [1 ft]; 500 µm mesh) by kicking the substrate so that disturbed invertebrates are swept into the net.

Labeling

Macroinvertebrate sample vials are double labeled. One external label is made using an electronic label maker, as for water chemistry and periphyton samples, but an additional internal paper label is also included. The backup paper label is necessary due to the ability of leaking ethanol to cause traditional labels to "bleed" the ink. The labels must include the following information: Stream Name, Stream code, Park code, date (yyyymmdd format), sample type, county/state, and **the number of vials in the sample**. Because the sample may not fit in one bottle, it is necessary to add "1 of 2" and "2 of 2." This prevents a laboratory technician at the contract lab from mistakenly processing only half of the sample. This information needs to be both on the internal and external label. The need to include the number of vials on the labels does not preclude the advance labeling of the bottles; the crew may affix the label and add a second label after sampling.

The paper label should be prepared on weather resistant paper (i.e., Rite-in-the-Rain) and filled out legibly in pencil (see premade example below).

Klamath Network, National Park Service Macroinvertebrates				
Park Code:				
Stream Name:				
Site Code:				
Date:				
County, State:				
of				

Collection Procedure

- 1. Starting downstream, begin collection at Transect A.
- 2. Determine a random starting point using the seconds on the watch (0-19 sec: Left; 20-39 sec: Center; 40-60 sec: Right).
- 3. If the flow at this spot is enough to fully extend the net, treat as a "riffle/run sample." If not, treat as a "pool/glide" sample (see #4).
 - a. In the section of river chosen using the random method, firmly plant the net down, with the opening facing upstream.
 - b. Holding the net firmly in place with your knees, start scrubbing the substrate in front of the net, in a 1 ft² area directly in front of the net opening. Visually define a square quadrat, using the net frame dimensions as a guide. Another useful trick is to measure the size of the wading shoe sole, to know if your shoe is approximately a "foot" long to guide the sampling.
 - c. Scrub the rocks within this quadrat using your hands or a scrub brush, until all invertebrates are dislodged from the rock. Place the rock aside and repeat until all rocks golf ball size or larger have been scrubbed.
 - d. Keeping the net in position, use your feet to vigorously kick the remaining substrate, dislodging any immovable rocks or any fine substrate. Kick for a 30 count.
 - e. Pull the net out of the water, and wash the collected bugs, detritus, and sediments into the bottom of the net.
 - f. Grabbing the bottom of the net, invert the net so that you can shake all collected material into a 5 gal bucket, partially filled with stream water.
 - i. If the net has a "dolphin bucket (Figure 2)," the contents can be washed into the bottom, the dolphin unscrewed, and emptied into the bucket. Immerse dolphin in the 5 gal bucket, and tap against the side to empty contents.

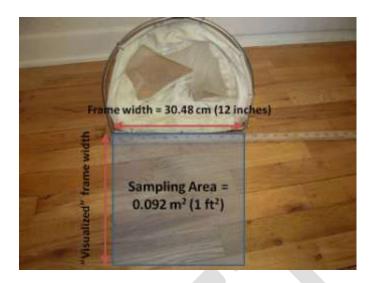


Figure 1. Example D-frame kicknet used for macroinvertebrate collection.



Figure 2. Dolphin bucket (top – attached; bottom – unscrewed for dumping) at end of invertebrate sampling net.

- g. **Fill out the data form**, indicating the habitat type (riffle or run) and the dominant substrate type (fine, gravel, or coarse).
- h. Proceed to the next transect.
- 4. If the sampling location is in a slow water habitat (pool or glide) that will not fully extend the net, use the alternate sampling technique.
 - a. Delineate a square foot, as above, using the dimensions of the net as a guide.
 - b. With the net in the water, pick up any larger substrates from within the quadrat and scrub the substrate, retaining the organisms within the net.
 - c. After all the large substrate has been scrubbed from the quadrat, actively sample the quadrat with the net and your feet. Use a single foot to disturb the quadrat while scrapping the area behind the foot with the net. Keep the net moving to keep swimming organisms trapped within the net. Actively sample this way for a 30 count.
 - d. Empty contents into the 5 gal bucket (the same one in which the detritus, bugs, etc. from previous transects were done).
 - e. **Fill out the data form**, indicating the habitat type (riffle or run) and the dominant substrate type (fine, gravel, or coarse).
 - f. Proceed to the next transect.
- 5. When all 11 transects have been sampled and data form filled out, process the sample to remove excess detritus and rocks.
 - a. Ensure that the 5 gal bucket containing the sample is approximately half full or more of stream water.
 - b. Try to remove large leaves, sticks, and leaves; vigorously shaking the leaves in the water to ensure no macroinvertebrates remain. Visibly inspect and gently remove any stubborn invertebrates with your finger. Generally, they will not bite.
 - c. Elutriate the sample to separate the heavier substrates (rocks, gravel, and sand) from the lighter detritus and invertebrates. Using two hands, gently swirl the contents of the bucket so that lighter material is lifted up (like panning for gold). Pour off the contents into a second 5 gal bucket. Repeat a minimum of five times; you can use less water each time, which helps swirl faster.
 - d. Sort through the remaining gravel, sand, and sediments for snails and rock-cased caddisflies (Trichoptera see below) that may remain. Pick any out and add to elutriate (the decanted material containing the invertebrates and lighter detritus) that was poured off.



- e. Pour elutriate through a $500 \mu m$ sieve. You can do this in batches; not all the material needs to be poured through at once.
- f. Once the material is on the screen, use a spoon to scoop the material and place in a 1 liter collection bottle. Continue pouring the sample through the sieve and transferring the material in the collection bottle until the entire sample has been processed.
- g. Two or more bottles may be necessary, depending on the amount of detritus and sediments still in the sample. Do not fill any individual bottle more than two-thirds full with the sample (adequate space is necessary for preserving the sample).
- h. Fill the collection bottle(s) with 95% Ethanol. Place paper label inside. Seal and invert several times to ensure that the Ethanol permeates the entire sample. Do not shake; this may damage the specimens, making identification difficult. Wrap bottle top with vinyl tape.
- i. Make sure the sample is properly labeled, all bottles are accounted for, and the data form is properly filled out.
- j. Rinse the net in the stream water, removing any lingering detritus.
- k. Samples should be transported back to the housing, but no special storage is needed.

Literature Cited

Peck, D. V., A. T. Herlihy, B. H. Hill, R. M. Hughes, P. R. Kaufmann, D. J. Klemm, J. M. Lazorchak, F. H. McCormick, S. A. Peterson, P. L. Ringold, *and others*. 2006. Environmental Monitoring and Assessment Program – Surface waters western pilot atudy: field operations manual for wadeable streams. U.S. Environmental Protection Agency, Office of Research and Development, Washington, D.C.